DEPMEDS LABORATORY PROCEDURES DEPARTMENT OF CLINICAL SUPPORT SERVICES U.S. ARMY MEDICAL DEPARTMENT CENTER AND SCHOOL FORT SAM HOUSTON, TEXAS 78234-6137

MCCS-HCL STANDING OPERATING PROCEDURE 01 November 01

ACTIVATED PARTIAL THROMBOPLASTIN TIME BY MLA ELECTRA 750

1. PRINCIPLE:

- a. The MLA Electra 750 utilizes a photometric detection technique to determine the clotting endpoint. When cephalin with activator and calcium chloride are added to anticoagulated plasma, a fibrin clot will form. The time it takes for the formation of this clot is the Activated Partial Thromboplastin Time (APTT).
- b. The APTT is a screening procedure used primarily to evaluate abnormalities in the enzymes and cofactors of the intrinsic coagulation pathway necessary for prothrombin activation.

2. SPECIMEN:

- a. Collect one part 3.8% (0.109 M) sodium citrate to 9 parts of whole blood in a plastic syringe or siliconized glass tube.
- b. Centrifuge within one hour after collection for approximately 15 minutes at 3300 RPM.
- c. Refrigerate at 2-8°C in a plastic or siliconized tube with stopper.
- d. Test plasma within 4 hours of draw. Specimens may be kept at room temperature if tested within 2 hours.
- e. For extended storage, rapidly freeze plasma at -20°C.

3. REAGENTS AND EQUIPMENT:

NOTE: Most reagents and controls are made of human or animal blood and tissue. Use of Personal Equipment (PPE) to prevent possible contamination or infection is required.

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- a. Actin activated cephaloplastin reagent.
 - (1) Store at 2-8°C.
 - (2) Reconstitute with purified water per manufacturer's instructions.
 - (3) After reconstitution store at 2-8°C; use within 24 hours.
 - (4) Record expiration date and time on label.
- b. Calcium chloride reagent 0.02 M.
- c. Controls.
 - (1) Store at 2-8°C.
 - (2) Reconstitute Normal and Abnormal with purified water per manufacturer's instructions
 - (3) After reconstitution store at 2-8°C, use within 24 hours
 - (4) Record expiration date and time on label.
- d. MLA Electra 750: Check Temperature and timer per MLA Initial Set-up S
- e. 0.2 mL pipet.
- f. 0.1 mL pipet.
- g. Pipet tips.
- h. MLA cuvettes.
- i. Purified water.

4. QUALITY CONTROL:

- a. Check instrument temperature, $37.2^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$.
 - (1) Record on Temperature Chart at beginning of shift.

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- (2) If temperature exceeds 37.7°C or falls below 36.7°C, notify supervisor and Medical Maintenance section.
- b. Normal and abnormal controls.
 - (1) Run at the start of each shift, at the beginning of each specimen run and when new reagent is used.
 - (2) Record all results on APTT QC chart.
 - (3) If any result exceeds ± 2 standard deviations, rerun control.
 - (a) If within range, record and continue run.
 - (b) If it exceeds a second time, record results and notify supervisor.

5. PROCEDURE:

- a. Turn on MLA 750. Allow a 5-minute warm-up period, or wait until AT TEMP indicator lights, whichever is longer.
- b. Check that LAMP LEVEL switch is at "B" (middle position).
- c. Prewarm Calcium Chloride in calcium reservoir for at least 5 minutes or until at operating temperature, 37°C.
- d. Set switch to APTT.
- e. Place two cuvettes in the heating block for each specimen required and add 0.1 mL of Actin to each.
- f. Add 0.1 mL of control or patient's plasma to each tube and mix.
- g. Set time for 5 minutes.
- h. After 5 minutes, place first cuvette to be tested in test station.
- i. Use 0.1 mL test pipet (blue top) to dispense 0.1 mL Calcium Chloride into cuvette holding Actin and test plasma.

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- j. Record test result.
- k. Repeat step "I" on duplicate specimen.

6. RESULTS:

- a. Agreement of results.
 - (1) Within 2 seconds.
 - (2) If results do not agree, run a second set of two cuvettes.
- b. Report the averaged APTT result to the nearest second along with the normal range.
 - c. Range.
 - (1) Normal: 24-35 seconds (Should be established by each laboratory.)
 - (2) Critical: Greater than 68 seconds.
 - d. Abnormal APTT results on patients who are not on anticoagulant therapy should be repeated.
 - e. If results are critical, notify physician immediately and document notification.

7. PROCEDURAL NOTES:

- a. Patient tube must be accurately filled; over or under filling can cause erroneous results.
- b. Ensure all reagents are well mixed and have stood for 15 minutes and are mixed again before testing.
- c. Allow 20 minutes for 10 mL of refrigerated reagent to come to analyzer temperature.
- d. Actin activated cephaloplastin reagent should not be held at 37°C longer than 60 minutes.

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- e. Use clean pipet tip on instrument pipette for each test to prevent carry over contamination.
- f. Insure that reagent is directed into the bottom of the tube and not run down the side of the cuvette. This provides proper mixing of the specimen and reagent.
- g. Patient plasma should not be incubated longer than 5 minutes.

8. LIMITATIONS:

- a. Operating temperature range: 10 to 32°C.
- b. Storage temperature range: -40 to 55°C.
- c. Operating humidity range: 0-60%, noncondensing.
- d. Storage humidity range: 0-90%, noncondensing.

9. REFERENCES:

- a. Brown, B.A., <u>Hematology: Principles and Procedures</u>. 6th ed., Philadelphia: Lea and Febiger, 1993.
- b. MLA Electra 750 Instruction Manual, Medical Laboratory Automation, Inc., 270 Marble Ave., Pleasantville, NY 10570-2982, 1990.